

CLAIMS

What is claimed is:

1. A recombinant vector comprising isolated DNA encoding a snRNA, wherein the isolated DNA comprises an insertion cassette contained between at least two
5 insertion sites.
2. The vector of Claim 1, wherein the snRNA is selected from the group of snRNAs with splicing functions.
3. The vector of Claim 2, wherein the snRNA is U1 snRNA.
4. The vector of Claim 2, wherein the snRNA is U6 snRNA.
- 10 5. The vector of Claim 3, wherein the insertion cassette comprises a modification fragment of about 30 base pairs of DNA.
6. The vector of Claim 5, wherein the modification fragment contains a modification to the first 11 nucleotides of a U1 snRNA.
7. The vector of Claim 6, wherein a single nucleotide is modified.
- 15 8. The vector of Claim 6, wherein a plurality of nucleotides are modified.
9. A recombinant vector comprising isolated DNA encoding a snRNA, wherein the isolated DNA comprises a Bae 1 restriction fragment.

10. The vector of Claim 9, wherein the isolated DNA further comprises two insertion sites formed by the excision of the Bae 1 restriction fragment.
11. The vector of Claim 10, wherein the two insertion sites comprise the complements of DNA sequences of SEQ ID NO: 2 and SEQ ID NO: 3.
- 5 12. The vector of Claim 10, further comprising an insertion cassette.
13. The vector of Claim 12, wherein the insertion cassette contains the same number of nucleotides as contained in the Bae 1 restriction fragment.
14. The vector of Claim 12, wherein the insertion cassette contains more nucleotides than were contained in the Bae 1 restriction fragment.
- 10 15. A method of producing a recombinant vector comprising isolated DNA encoding a product of interest, wherein the isolated DNA comprises an insertion cassette contained between at least two insertion sites comprising the following steps:
- 15 (a) inserting isolated DNA encoding a product of interest into the vector;
- (b) contacting the isolated DNA with a dual cleavage restriction enzyme that excises a restriction fragment comprising a double-stranded DNA modification fragment linked to a single-stranded DNA overhang at each end of the modification fragment;
- 20 (c) excising the restriction fragment from the isolated DNA of the vector such that at least two insertion sites are formed in the isolated DNA;
- (d) obtaining an insertion cassette wherein the single-stranded overhang at each end of the modification fragment comprises a

DNA sequence complementary to the DNA sequence of the insertion sites formed in the isolated DNA; and

- (e) ligating the insertion cassette into the isolated DNA of the vector between the insertion sites formed by the excision of the restriction fragment,

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thereby producing a recombinant vector comprising an insertion cassette contained between at least two insertion sites.

16. The method of Claim 15, wherein the product of interest is selected from the group of snRNAs with splicing functions.

- 10 17. The method of Claim 15, wherein the snRNA is U1 snRNA.

18. The method of Claim 15, wherein the snRNA is U6 snRNA.

19. The method of Claim 17, wherein the insertion cassette comprises a modification fragment of about 30 base pairs of DNA.

- 15 20. The method of Claim 19, wherein the insertion cassette contains a modification to the first 11 nucleotides of a U1 snRNA.

21. The method of Claim 20, wherein a single nucleotide is modified.

22. The method of Claim 20, wherein a plurality of nucleotides are modified.

23. The method of Claim 17, wherein the dual cleavage restriction enzyme is Bae 1.

- 20 24. The method of Claim 17, wherein each insertion site consists of about 5 nucleotides.

25. A cell transformed by a recombinant vector comprising isolated DNA encoding a snRNA, wherein the DNA comprises an insertion cassette contained between at least two insertion sites.
- 5 26. The cell of Claim 25, wherein the cell is a mammalian cell.
27. A cell library comprising cells transformed by a plurality of recombinant vectors comprising isolated DNA encoding a snRNA, wherein the DNA comprises an insertion cassette contained between at least two insertion sites.
- 10 28. The cell library of Claim 27 comprising insertion cassettes containing at least about 10 different modification fragments.
29. A method of identifying a modification of a snRNA which suppresses transcription of a transcription product in a cell comprising:
- 15 (a) determining a base level of transcription of a transcription product in a cell;
- (b) producing at least 10 recombinant vectors comprising isolated DNA encoding a snRNA, wherein the isolated DNA of each of the recombinant vectors comprises an insertion cassette containing a different modification fragment contained between at least two insertion sites of the vector;
- 20 (c) introducing each vector containing a modification into a cell, under conditions suitable for delivery of the snRNA into the cell;
- (d) comparing the level of transcription of the transcription product in each cell containing a vector containing the modified snRNA with the base level of transcription of the transcription product in the cell; and
- 25 (e) determining which snRNA modifications inhibit transcription in the cell,

whereby, if the level of transcription of the transcription product in the cell containing the vector containing the modified snRNA is less than the base level of transcription of the transcription product in the cell, a modification which suppress expression of a transcription product in the cell has been identified.

- 5 30. A method of suppressing expression of a transcription product in a cell comprising:
- (a) producing a recombinant vector comprising isolated DNA encoding a snRNA, wherein the isolated DNA comprises an insertion cassette contained between at least two insertion sites;
- 10 (b) introducing the vector into the cell, under conditions suitable for delivery of the snRNA into the cell; and
- (c) utilizing the snRNA to inhibit transcription in the cell,
- thereby, suppressing expression of a transcription product in the cell.

- 15 31. A method of delivering an antisense targeting sequence into a cell nucleus comprising:
- (a) inserting an antisense targeting sequence into a recombinant vector comprising an isolated DNA encoding a snRNA, wherein the DNA comprises an insertion cassette contained between at least two insertion sites; and
- 20 (b) introducing the vector into the cell, under conditions suitable for delivery of the antisense targeting sequence across the cell membrane and into the cell nucleus,

whereby the antisense targeting sequence is delivered to the cell nucleus.